**Contrast enhanced X-Ray MicroCT scans of fruit**

Zi Wang¹, Pieter Verboven¹, Bart Nicolai¹,²

¹MeBioS, Department of Biosystems, KU Leuven, Willem de Croylaan 42, 3001 Leuven, Belgium
Email: zi.wang@kuleuven.be, pieter.verboven@kuleuven.be

²Flanders Centre of Postharvest Technology, Willem de Croylaan 42, 3001 Leuven, Belgium
Email: bart.nicolai@kuleuven.be

**Abstract**
Incremental improvements in the quality control and preservation of high value fruit products is increasingly reliant on computer modelling and simulations. To obtain representative 3D models of biological tissue, the collected imaging data must be accurate. Although methods exist to capture data in 3 dimensions, none compare to X-ray micro-computed tomography (μCT) in simultaneous throughput and quality. However, because of the high energy nature of X-Ray photons, soft biological materials with similar atomic composition cannot be easily distinguished from each other, resulting in geometric inaccuracies in the derived 3D models. To combat this problem, we have adapted radio-opaque contrast enhancing solutions to mark intercellular boundaries of fruit cortex tissue. Cortex samples from apples and pears were excised and immersed within a cesium iodide contrast solution for up to 2 hours, and were subsequently scanned at a resolution of 3μm. The resulting images show distinctly labelled intercellular boundaries, which helps to improve the image segmentation process and accuracy of the derived 3D models.

**Keywords**: Contrast agents, microCT, fruit tissue.

**1 Contrast enhancement of fruit cortex tissue**

Previous studies on microCT scans of apple cortex tissue relied on the watershed segmentation method to mark cellular boundaries for binarization of the greyscale image [1]. However, this method is limited to highly aerated tissue as denser cell clusters lack the necessary number of local minima for the technique to work effectively. By embedding contrast agents within the cortex tissue, segmentation can be done more accurately as the intercellular boundaries can be differentiated by the increased greyscale intensity in the reconstructed image.

![Representative images of Kanzi apple microCT scans without and with contrast enhancement (top and bottom images respectively), along with the color labelled images after segmentation.](image-url)
Figure 1 demonstrates the value of utilizing a 10% cesium iodide (w/v) contrast enhancing solution to label cellular boundaries in apple cortex tissue. When a non-enhanced scan is compared with the contrast enhanced image, localization of cell boundaries is uncertain in the former, but quite clear in the latter. When the reconstructed images are binarized and color labelled, it is obvious that the enhanced images allow for a greater degree of object separation, whereas a significant number of unenhanced cell clusters are inseparable with only watershed segmentation incorporated into the workflow.

2 Assessing sample integrity following contrast delivery
As apple cortex tissue has a relatively high air fraction, it is possible to assess whether embedding the contrast solution has any adverse effects on the sample being scanned. Since the contrast agents now occupy many of the previous airspaces, semi-automatic image registration was done on the dataset to determine whether these air pockets exhibit any notable distortions resulting from either the contrast solution used or the delivery method itself. Although superimposed datasets exhibit single digit pixel deviation in the alignment which can be attributed to the semi-automatic registration protocol used, no notable distortions can be observed in the volume of interest extracted for analysis.

3 Transferability testing
Pear cortex tissue differ significantly from apples, as the parenchyma cells are smaller and the void network occupy a reduced volume fraction compared to that of apple tissue [2]. These differences combined with the limitations of microCT imaging typically results in large numbers of inseparable cell clusters. By introducing the contrast solution to excised pear cortex tissue, a notable enhancement was indeed observed where the cell boundaries are marked with the contrast solution. The effect was especially pronounced around stone cells, where boundary localization was previously not possible without the use of phase contrast synchrotron X-Ray CT [2].

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References